Motility

Given the different viscosity of the medium and the placement of the colony on/in the medium, it is possible to test for presence or absent of twitching, swarming and swimming.

Three kinds of plates were prepared each containing a different agar concentration, see table 9 for swimming, table 10 for twitching and table 11 for swarming. The concentrations were 1.5 % for twitching, 0.53 for swarming and 0.3 % for swimming. All plates were left at room temperature until inoculation the following day.

Strains were grown ON on LB plates and a single colony was transferred with a toothpick to a motility plate. For twitching the colony was stabbed through the agar to the bottom of the media, for swarming the colony was placed on the surface on the media, while for swimming the surface was pierced and the colony deposited in the middle of the agar. Swarming and swimming plates were incubated at 30 °C for 24 hours, while twitching plates were incubated at 37°C for 48 hours. In twitching PAO1 *pilA* were used as a neative control and in swimming and swarming PAO1 *fliF* were used as a negative control.

Table 9: Assay for swimming motility plate. Yield: two plates with 0,3 % agar. (dry the plates for 15min at 37C before use)

8 ml melted ABT medium (mix of A10 and BT medium) with 2 % agar
42 ml ABT preheated to 50 °C
1,25 ml 20 % glucose
1,25 ml 20 % Cas-amino acids

Table 10: Assay for twitching motility plate. Yield: one plate with 1,5 % agar (dry the plates for 15 min at 37C before use)

19 ml melted ABT medium (mix of A10 and BT medium) with 2 % agar
6 ml ABT preheated to 50 °C
0,625 ml 20 % glucose
0,625 ml 20 % Cas-amino acids

Table 11: Assay for swarming motility plate. Yield: two plates with 0,53 % agar. (dry the plates for 5min at 37C before use)

14 ml melted ABT medium (mix of A10 and BT medium) with 2 % agar
36 ml ABT preheated to 50 °C
1,25 ml 20 % glucose
1,25 ml 20 % Cas-amino acids